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Identification of mec*A* genes in environmental *Staphylococcal* isolates

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Background: In the past Hospital isolates of *Staphylococcus aureus* (MRSA) were considered more drug resistant than community acquired strains. However community acquired MRSA strains have now have developed drug resistance commensurate to Hospital strains. The question arise as to what environmental factors have led to this development, is it just the widespread use of antibiotics in the non-hospital environment or are there potentially other factors. Recently, increased attention has been paid to multidrug-resistant coagulase-negative Staphylococci (MRCNS) in hospitals. These strains are recognized as opportunistic pathogens in the immuno- suppressed and hospital strains are also a potential reservoir for *mecA* genes. This current study was to investigate antibiotic resistance in Staphylococci isolated from a non-hospital environment in common use by humans, in this case from public washrooms.

Methods: Swabs taken from public washrooms were analysed. Staphylococci were identified using the Bruker MALDI-TOF biotyper system. The minimum inhibitory concentrations (MIC) of Staphylococcal isolates against 30 antibiotics, including oxacillin, erythromycin, amoxicillin, and vancomycin were determined using the MicroScan Walkway 96 plus automated system (Simens Healthcare Diagnostics, CA, USA). For methicillin resistant Staphylococcal isolates, a rapid latex agglutination assay kit was used to determine PBP2' according the manufacturer's instructions (Oxoid, Basingtoke, UK). Genomic DNA of the isolates was prepared and *mec* complex was determined by polymerase chain reaction (PCR).

Results: 33% of Staphylococcal isolates (both coagulase-positive and coagulase-negative) were multidrug resistant, including non-β-lactam antibiotics such as fusidic acid, gentamycin, erythromycin, chloramphenol. MICs for oxacillin varied from 0.75 to 64 mg/l. The presence of mec*A* was confirmed in all MRSA and MRCNS isolates. Moreover, we determined *mec* complexes were widely spread between isolates and represented all classes (A, B,C,C1,D) of *mec* complexes.

Conclusion: We have demonstrated that non-healthcare establishments such as washrooms are potential reservoir for antibiotic resistant Staphylococci. Using combination of PBP2' agglutination test and PCR methods we have shown that all methicillin resistant environmental *staphylococci* isolates carried *mecA*. Such a widespread presence of drug resistance determinants in these human environments is a potential source to aid the spread, sustainability and development of drug-resistance both in hospitals and community strains.

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Staphylococcal cassette chroromosome mec (SCCmec) typing of methicillin-resistant *Staphylococcus aureus* (MRSA) isolates from patients attending Tengku Ampuan Afzan hospital (HTAA) in Kuantan, Pahang, Malaysia

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Background: The adaptive power of *Staphylococcus aureus* to antibiotics lead to the emergence of methicillin-resistant *S. aureus* (MRSA) in the early 1960s. The cause of resistance to methicillin is the acquisition of the *mecA* gene, situated on a mobile genetic element called the staphylococcal cassette chromosome mec (SCCmec).

Methods: One of the most important techniques used to investigate the molecular epidemiology of *S. aureus* is SCCmec typing. This technique has been used to study the evolution of the MRSA and to study their subsequent worldwide dissemination. Twenty-eight MRSA isolates were subjected toSCCmec typing by duplex real time PCR. The MRSAs were isolated by the Bacteriology Laboratory in Hospital Tengku Ampuan Afzan (HTAA), Kuantan, Pahang, Malaysia from inpatients admitted during the period from 1stApril to 30th September, 2010. The MRSA isolates were re-identified by known bacteriological methods and the minimal inhibitory concentration (MIC) of oxacillin was determined by E-test. The antibiotic susceptibility was tested, by disc diffusion method to 7 different antibiotics.

Results: Four major sites were listed (Fig 3): skin and soft tissue infections 14 (50%), ENT infections 7 (25%), respiratory tract infections 1 (3.57%), blood stream infections 5 (17.85%), and other body fluids 1 (3.57%).

Resistance to oxacillin was 100%, with an MIC >4 μg/mL. Resistance to other antibacterial drugs was erythromycin 82.1%, gentamicin 75%, tetracycline 78.6%, and trimethoprim-sulfamethoxazole 78.6%. None of the isolates was resistant to vancomycin or chloramphenicol. 78.5% (22/28) were shown to be of SCCmec-type III and 21.5% (6/28) were of type IV.

Conclusion: The results confirm observations in several other neighboring Far Eastern countries and corroborates the epidemicity of these two SCCmec types in Kuantan, Malaysia.

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